

Nicotinamide effects on the metabolism of human fibroblasts and keratinocytes assessed by quantitative, label-free fluorescence imaging: supplement

ZHIYI LIU,^{1,3} CHUNG-YI CHIANG,² JOHN NIP,² LIN FENG,² YANG ZHANG,¹ SHEILA ROCHA,² AND IRENE GEORGAKOUDI^{1,*} 

¹*Department of Biomedical Engineering, Tufts University, Medford, MA 02155, USA*

²*Unilever Research and Development, Trumbull, CT 06611, USA*

³*Currently with the State Key Laboratory of Modern Optical Instrumentation, College of Optical Science and Engineering; International Research Center for Advanced Photonics, Zhejiang University, Hangzhou, Zhejiang 310027, China*

**Irene.Georgakoudi@tufts.edu*

This supplement published with Optica Publishing Group on 20 September 2021 by The Authors under the terms of the [Creative Commons Attribution 4.0 License](https://creativecommons.org/licenses/by/4.0/) in the format provided by the authors and unedited. Further distribution of this work must maintain attribution to the author(s) and the published article's title, journal citation, and DOI.

Supplement DOI: <https://doi.org/10.6084/m9.figshare.15261843>

Parent Article DOI: <https://doi.org/10.1364/BOE.432561>

Supplementary information

for

**Nicotinamide effects on metabolism of human fibroblasts
and keratinocytes assessed by quantitative, label-free
fluorescence imaging**

Zhiyi Liu,^{1,3} Chung-Yi Chiang,² John Nip,² Lin Feng,² Yang Zhang,¹ Sheila Rocha,² and
Irene Georgakoudi^{1,*}

¹*Department of Biomedical Engineering, Tufts University, Medford, MA 02155, USA*

²*Unilever Research and Development, Trumbull, CT 06611, USA*

³*Currently with the State Key Laboratory of Modern Optical Instrumentation, College of Optical Science
and Engineering; International Research Center for Advanced Photonics, Zhejiang University, Hangzhou,
Zhejiang 310027, China*

**Irene.Georgakoudi@tufts.edu*

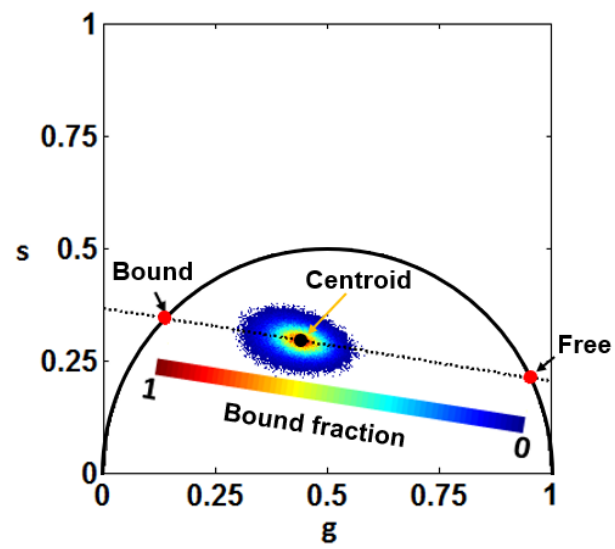


Fig. S1. Phasor plot of an NAD(P)H image, according to the fluorescence lifetime at each pixel within the cytoplasm area.

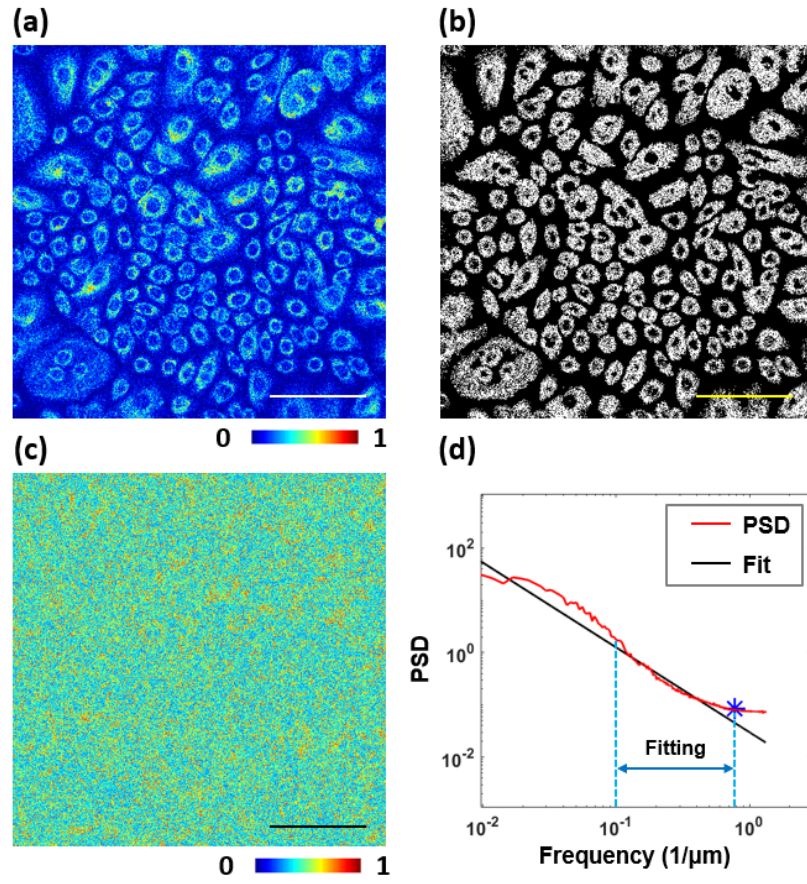


Fig. S2. Schematic showing the analysis of mitochondrial clustering. (a) Raw NAD(P)H intensity image. (b) Binary mask with cytoplasmic area only, excluding nuclear and background areas. (c) Clone stamped image of the NAD(P)H intensity signals within the binary mask. (d) PSD of the clone stamped image along with inverse power law expression fit. Scale bar: 100 μm.

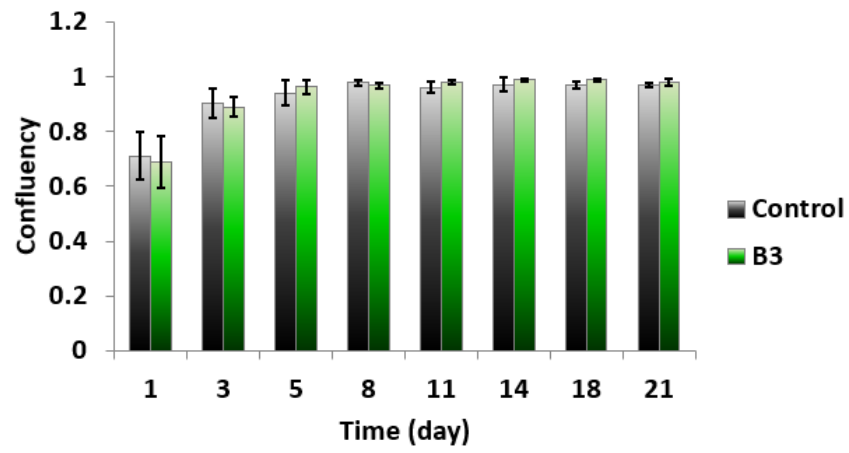


Fig. S3. Mean and standard deviation of fibroblast confluency at different time points in response to B3 supplementation.

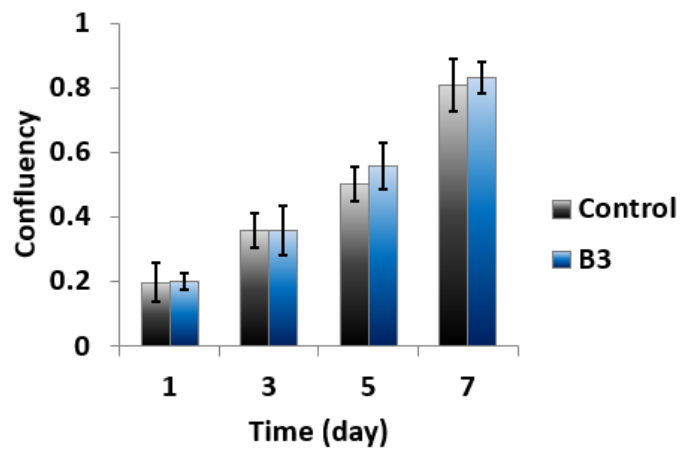


Fig. S4. Mean and standard deviation of keratinocyte confluency at different time points in response to B3 supplementation.

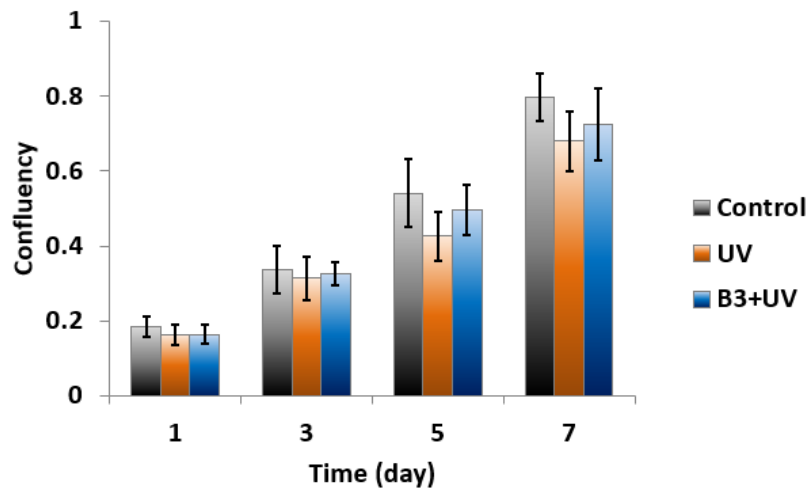


Fig. S5. Mean and standard deviation of keratinocyte confluency at different time points in response to B3 supplementation and UV irradiation (from Day 4 to Day 6).

Table S1. Comparison of optical biomarkers at different time points relative to Day 1 for the fibroblast study. *NS*: no significance; *, $p < 0.05$.

Biomarker	Treatment	3	5	8	11	14	18	21
Redox ratio	Control	<i>NS</i>	<i>NS</i>	*	*	*	*	*
	B3	<i>NS</i>	<i>NS</i>	<i>NS</i>	*	*	*	*
NAD(P)H bound fraction	Control	*	*	*	*	*	<i>NS</i>	<i>NS</i>
	B3	<i>NS</i>	<i>NS</i>	*	*	*	*	*
Mitochondrial clustering	Control	<i>NS</i>	<i>NS</i>	*	*	*	*	*
	B3	<i>NS</i>	<i>NS</i>	<i>NS</i>	*	<i>NS</i>	<i>NS</i>	<i>NS</i>

Table S2. Comparison of optical biomarkers at different time points relative to Day 1 for the keratinocyte study without UV exposure. *NS*: no significance; *, $p < 0.05$.

Biomarker	Treatment	3	5	7
Redox ratio	Control	<i>NS</i>	*	*
	B3	<i>NS</i>	*	*
NAD(P)H bound fraction	Control	<i>NS</i>	<i>NS</i>	<i>NS</i>
	B3	<i>NS</i>	<i>NS</i>	*
Mitochondrial clustering	Control	*	*	*
	B3	*	<i>NS</i>	<i>NS</i>

Table S3. Comparison of optical biomarkers at different time points relative to Day 1 for the

keratinocyte study with UV exposure. *NS*: no significance; *, $p < 0.05$.

Biomarker	Treatment	3	5	7
Redox ratio	Control	<i>NS</i>	*	*
	UV	*	<i>NS</i>	*
	B3+UV	<i>NS</i>	<i>NS</i>	<i>NS</i>
NAD(P)H bound fraction	Control	<i>NS</i>	<i>NS</i>	<i>NS</i>
	UV	<i>NS</i>	*	*
	B3+UV	<i>NS</i>	*	*
Mitochondrial clustering	Control	<i>NS</i>	<i>NS</i>	<i>NS</i>
	UV	<i>NS</i>	*	*
	B3+UV	<i>NS</i>	<i>NS</i>	<i>NS</i>

Table S4. Assessing differences in each optical biomarker among different treatments at distinct time points for the keratinocyte study with UV exposure. *NS*: no significance; *, $p < 0.05$.

Biomarker	Treatment	1	3	5	7
Redox ratio	Control vs. UV	<i>NS</i>	<i>NS</i>	*	*
	UV vs. B3+UV	<i>NS</i>	<i>NS</i>	*	<i>NS</i>
	Control vs. B3+UV	<i>NS</i>	<i>NS</i>	*	*
NAD(P)H bound fraction	Control vs. UV	<i>NS</i>	<i>NS</i>	*	<i>NS</i>
	UV vs. B3+UV	<i>NS</i>	<i>NS</i>	<i>NS</i>	<i>NS</i>
	Control vs. B3+UV	<i>NS</i>	<i>NS</i>	*	*
Mitochondrial clustering	Control vs. UV	<i>NS</i>	<i>NS</i>	*	*
	UV vs. B3+UV	<i>NS</i>	<i>NS</i>	<i>NS</i>	*
	Control vs. B3+UV	<i>NS</i>	<i>NS</i>	<i>NS</i>	<i>NS</i>